
EVIDENCE THAT INITIATION FACTOR 1 INTERACTS WITH 23S RIBOSOMAL RNA AT THE HIGHLY CONSERVED ALPHA SARCEN SITE ^{MAS}

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The cleavage reaction catalyzed by the cytotoxic protein alpha-sarcin on 23s rRNA can be duplicated by using a DNA probe complementary to the region targeted by the toxin (2646-2574). Previous studies on this site utilizing RNase H generate nucleic acid cleavage during *in vitro* poly-Phe synthesis. Here we use a metal chelator, 1,10-phenanthroline in the presence of Cu⁺⁺ ions and mercaptopropionic acid to

cleave the nucleic acid. We found that cleavage at 2655-2667 and 2654-2664 is possible only with the addition of a crude S150 fraction. Further studies showed that IF-1 was the determining factor involved in generating this cleavage. These results suggest that IF-1 may play a role in producing conformational changes in the alpha-sarcin region of the 23s rRNA.

PHARMACOLOGY AND TOXICOLOGY

THE EFFECTS OF TAXOL AND STRESS RESPONSE/ HEAT SHOCK IN MULTIPLE DRUG RESISTANT C-6 GLIOMA ^{MAS}

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Multidrug-resistant cells are thought to maintain low intracellular cytotoxic drug concentrations through the active efflux of drugs across the cell membrane. P-glycoprotein, encoded by the MDR1 gene, is believed to mediate this energy-dependent drug efflux by interacting directly with various amphipathic compounds. We have isolated a multidrug resistant strain of rat C-6 Glioma by exposing cells to progressively increasing concentrations of colchicine-a microtubule destabilizing compound that selectively kills MDR negative cells. Drug resistance was verified by determining ED₅₀ values of both the parent (82.0 ± 23.5 ng/ml) and the drug resistant strain (1490.3 ± 157.7 ng/ml) upon exposure to increasing concentrations of taxola microtubule

stabilizing agent. The drug resistant strain also exhibited a significant decrease ($p < 0.05$) in the rate of growth (cells/ml/24 hours) in comparison to the parent strain. To establish a correlation between multiple drug resistance and the stress response, both strains were subjected to heat shock at 42°C for 30 minutes. The presence of heat shock proteins (hsp 70, hsp 90, hsp 32) was assessed by SDS-PAGE; no significant differences were found in either of the treated and untreated parent and drug resistant strains. Protein concentrations were significantly increased in the heat treated and untreated drug resistant strains. In addition, glutamine synthetase (GS), cyclic nucleotide phosphohydrolase (CNP), and heme

oxygenase (HO) enzyme activities were significantly increased in both heat treated and untreated drug resistant strains. Exposure of both the parent and drug resistant strains to varying concentrations of verapamil, a substrate for p glycoprotein, was used to show resistance modification to taxol.

The results suggest that there is a correlation between the stress response to heat shock and multiple drug resistance. They also suggest that C-6 Glioma would be a model system for testing other antineoplastic drugs and other resistance modifying agents.

EFFECTS OF ALLOXAN-INDUCED DIABETES (TYPE I) AND HYPERCHOLESTEROLEMIA ON THE CARDIOVASCULAR SYSTEM OF ADULT MALE RABBITS ^{MAS}

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The modified Langendorff rabbit heart preparation was used to assess the ability of diabetic and non-diabetic hearts to withstand the stress of 20 minutes of global ischemia followed by 30 minutes of reperfusion (ischemia/reperfusion injury). Cylindrical strips of aortas were used in 25-ml tissue baths to determine the influences of diabetes (alloxan at 140 mg/kg i.v.), aminoguanidine (15 mg/kg/day p.o.), and hypercholesterolemia (3% coconut oil, 3% peanut oil and 0.5% cholesterol) on voltage-dependent and adrenergic receptor-dependent vascular contraction and on endothelium-dependent and endothelium-independent relaxation. Cardiac functionality was evaluated by measuring double products (DP), heart rates (HR), left ventricular developed pressures (LVDP), and left ventricular systolic pressures (LVSP) immediately

before ischemia (preischemia) and 30 minutes after terminating global ischemia (postischemia). Preliminary data indicate that diabetic hearts regardless of pretreatment tended to start with poorer cardiac performance, but lost a smaller percentage of functionality as a result of global ischemia. Hypercholesterolemia of 4 months duration failed to decrease cardiac performance or to increase myocardial sensitivity to ischemia/reperfusion (I/R) injury. Aminoguanidine, known to prevent the accumulation of advanced glycated end products, did not protect hearts against I/R injury. Aminoguanidine increased the vascular reactivity of aortic strips in the presence of 10 M⁻⁵ norepinephrine, but not in the presence of KCl (40 mM). The ability of aortas to relax was normal across all treatments.

POTENTIAL OF Y79 RETINOBLASTOMA AS A MODEL DRUG SCREENING SYSTEM ^{MAS}

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Y79 is a human retinoblastoma cell culture line grown optimally in

suspensions but also grown in monolayer culture when the growth

surface is pre-treated with attachment factors such as poly D-lysine or fibronectin. Y79 shows many characteristics of normal retinal cells including the presence of enzymes responsible for melatonin synthesis. Both N-acetyltransferase (NAT), which catalyzes the conversion of serotonin to N-acetyl- serotonin, and hydroxyindole-O-methyltransferase (HIOMT), which catalyzes the conversion of N-acetylserotonin to melatonin, are active in this culture system. In our attempts to better understand migraine headache and to build superior anti-migraine drugs, we have been attracted to melatonin because light is a regulatory parameter for both melatonin and migraine. One of the chief problems in anti-migraine drug development has been a reliable, relatively straight forward system for evaluating the potential of new drugs. The physiological characteristics of Y79 in concert with the ease of culturing and treating the cells suggest that it has potential as a model in vitro system for screening anti-migraine drugs. In the preliminary studies reported here, Y79 cells were grown in suspension and in some cases in monolayer, in the presence of RPMI 1640 medium

containing 10% fetal calf serum. Cultures were incubated in a humidified 5% CO₂ atmosphere at 37 ° C. Drug treatments were for 24 hours, after which cells were harvested and homogenized by sonication. NAT activity was determined by incubation of homogenates at 30 ° C for 30 minutes in the presence of tryptamine and 14C-acetyl Co-enzyme A. Radiolabeled product was quantified by liquid scintillation spectroscopy and protein was measured by the Bio-Rad procedure. Current anti-migraine drugs were tested for activity in the system. Propranolol and sodium valproate both elevated NAT activity while the calcium channel antagonist flunarizine lowered activity. Dihydroergotamine and buspirone, a 5-HT_{1a} partial agonist both were without activity. Additionally, the 5-HT_{1a} agonist 8-OH-DPAT was used to test for the presence of receptor, and a dose-response curve was obtained with a K_d of approximately 1 nM. These pilot studies warrant further exploration of this system, and hint that the system may be predictive of prophylactic anti-migraine drug activity if not acute activity. Serotonin receptors may be present, but melatonin receptors have not been evaluated at this point.

RETINOBLASTOMA ^{MAS}

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Retinoblastoma, the eye cancer that affects the youth of our society, is a complex disease. Although the incidence of retinoblastoma has dropped dramatically since the turn of the century, it still strikes young, innocent children all too often. Early diagnosis of this disease is crucial for a promising prognosis. The two forms of retinoblastoma recognized are unilateral and bilateral retinoblastoma. Unilateral

disease affects only one of the child's eyes, whereas bilateral disease means that cancer has been found in both eyes. Retinoblastoma can develop as a result of an inherited gene abnormality, or it can occur spontaneously. Signs such as leukokoria, strabismus, esotropia or exotropia, and heterochromia usually prompt the testing for retinoblastoma. Diagnostic tests used to detect retinoblastoma are ultrasonography, CT

scans, and ophthalmoscopic examinations. The therapy implemented depends on the stage of the cancer at the time of diagnosis. Stages are classified as intraocular, extraocular, or recurrent. The three most common treatments are surgery, which may include enucleation, cryosurgery, or photocoagulation, radiation, and chemotherapy. The prognosis, again, depends on the stage

of retinoblastoma when the treatment was implemented. Follow-up exams are extremely important in order to watch for other cancers that may develop as a result of the gene abnormality or the treatment that was used for retinoblastoma. These cancers are referred to as second nonocular tumors and the most common form is osteosarcoma.

SOCIAL SCIENCES

WHAT CITIZENS DON'T KNOW CAN HURT THEM ^{AFS}

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Back in grade school I recall hearing the phrase, "Ignorance of the law is no excuse." The assumption was that by our knowledge of the law we would only knowingly commit crime, and in turn know if we were victims of crime. Though our understanding of law is most often common sense, i.e., we don't steal things, we don't assault others, etc., there are some laws which defy common sense. When Noranda/Crown Butte began exploration at Cooke city, Montana, the idea of a large gold mine did not seem possible. After all, who

would put a huge industrial complex on the border of the world's first national park? The General Mining Law of 1872 declares, "hard rock mining is the highest use of public lands" and hard rock miners like Noranda/Crown Butte have the "exclusive and preemptive right to mine." Most people don't know about this law. When people become aware of it, almost overwhelming disdain and outrage at its present day absurdity is the reaction. This law should be reformed.

PUBLIC ATTITUDES TOWARD MULE DEER IN AN URBAN ENVIRONMENT IN EASTERN MONTANA ^{TWS}

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As part of a broad study to develop management strategies for mule deer (*Odocoileus hemionus*) inhabiting strip mine reclamation and urban habitats in the vicinity of Colstrip, Montana, an effort was made to document, via mail survey, public attitudes toward deer

inhabiting the urban area. Deer-human conflicts in Colstrip have increased during recent years as an increasing number of deer have utilized forage resources, including shrubs and gardens, in the city. Results indicate that although deer are causing